NOXIOUS TOADS AND FROGS OF SOUTH AFRICA

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The major defence mechanism in frogs is via the secretion of toxins from their skin. In humans, intoxication may occur when part of the amphibian integument is ingested, as in the form of herbal medicines. Two groups of South African frogs have skin secretions that are potentially lethal to humans and animals. Toads (*Bufo* and *Schismaderma* species), the amphibians with which man and his pets most frequently have contact, secrete potent toxins with cardiac glycoside activity. Topical and systemic intoxication, while seen in humans, remains predominantly a veterinary problem. Intoxication by the red-banded rubber frog, which secretes an unidentified cardiotoxin, is far less common. The probable mechanisms of intoxication and management of a poisoned patient are discussed.

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Reports of human and veterinary toxicity caused by amphibians are not common in the literature.[1-4] However, little has been published on noxious South African amphibians, probably because the occurrence of toxicity is low and/or because toxicity has previously not been diagnosed. Anurans (frogs and toads) are the only amphibians indigenous to South Africa. *Bufo* (toads), *Schismaderma* (red toads) and *Phrynomantis* (rubber frogs) are the only genera of toxicological importance. The highly toxic tropical dendrobatid frogs used for dart poison[5] do not occur in South Africa. Toxicity remains primarily a veterinary problem. However, in view of the fact that humans have considerable contact with amphibians, particularly toads, physicians should have some knowledge of their toxicity.

Only skin secretions of amphibians are considered poisonous, although venom has been isolated from the plasma and eggs of some toads.[6,7] The amphibian skin houses mucous and granular ('poison') glands. Mucous glands are dispersed all over the body, whereas granular glands are localised in special regions, such as the paratoid poison glands behind the head in toads. Because of their locality the paratoid glands, which do not secrete saliva, are often erroneously referred to as 'parotid' glands. Granular glands produce a thicker, more toxic secretion. However, secretion from both mucous and granular glands may be poisonous.[8] While nearly all amphibians have a trace of toxin in their skin,[9] this is not equally developed in the different genera. Both gland types are under the control of sympathetic nerves and discharge following a variety of stimuli.[10] Mucous glands secrete mainly when stimulated by dryness, whereas granular glands require pressure, injury or any stress to the animal to cause secretion.[11,12] With some exceptions, notably *Bufo marinus* (not a South African toad), pre-metamorphic larvae lack toxic properties as granular glands develop only at metamorphosis.[13]

Amphibians are not equipped for speed, nor do they have any armament; as such they rely predominantly on chemical defence to deter predators.[14-18] As well as being unpalatable, the sticky, viscous properties of secretions may confer a protective advantage. This is seen in some species of *Breviceps* (rain frog),[19] whose defensive white secretions have earned it the name 'melk padda'. Such sticky toad secretions have given rise to the false superstition that handling of toads causes warts. Poisoning may cause local and/or systemic toxicity. In domestic and wild animals poisoning usually occurs following mouthing or ingestion of the amphibian. In humans, poisoning is likely to occur when secretions are brought into contact with skin, mucosa or conjunctiva.[20-22] Absorption of venom directly into the circulation may occur where cuts or abrasions are present.[7,23] Intoxication may also occur when part of the amphibian integument is ingested, as in the form of herbal medicines,[24] or when saliva containing toxins is inspired.[25] Systemic toxicity following topical application of venom to intact skin has not been documented. However, anuran venom introduced intradermally, rather than topically, causes a more marked and painful dermal reaction.[26] The fascinating practice of smoking dried toad skin and licking toads for their hallucinogenic effects has also been reported.[27] No amphibian causes actual mechanical trauma, except for the bullfrog (*Pyxicephalus*) which may inflict a nasty bite[28] with its teeth-like odontoids on the lower jaw.

BIOCHEMISTRY AND PHARMACOLOGY

Amphibians secrete a wide range of substances belonging to many chemical classes with varied pharmacological activities. Many of these compounds could serve the frog in defence as chemical irritants[29,30] secondary to a physiological role.[31,32] They include biogenic amines (e.g. catecholamines), various peptides (e.g. bradykinin, physalaemin and caerulein), indolealkylamines (e.g. serotonin), bufadienolides (one of the cardiac aglycones) and alkaloids.[33-36] There are cardiotoxins,

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neurotoxins, sympathomimetics, pressor agents, local anaesthetics and hallucinogens present in frog secretions.[27,40-42] The indolealkylamines (bufotenines or related tryptamine alkaloids) exert primarily oxytocic and pressor effects and are responsible for causing hallucinations.[27] Peptides from skin secretions of the South African platanna (*Xenopus laevis*) have even shown similarity to snake venom toxins.[43] Secretions from *Xenopus* makes it unpalatable as prey to other animals and also toxic to other frogs in contact with it. Intoxication of mammals by *Xenopus*, however, is not known (W D Haacke — personal communication). At high enough dosages all these compounds may be poisonous. Their equivalents in mammalian tissues occur in lower concentrations and are often less active.[44,45] For example, the amount of adrenaline stored in the paratoid glands of *Bufo gutturalis* (formerly *B. regularis*,[46] namely 10.7 mg per toad, is equivalent to the amount of adrenaline found in the adrenals of sheep, and greater than that found in man.[47]

When isolated from toad-skin secretions the cardio-active bufadienolide steroid aglycones (bufagins) and their derivatives, the bufotoxins, are structurally analogous to the well-known plant bufadienolide and cardenolide cardiac glycosides, such as those respectively found in medicinally applied squill (plant of the lily family) and digitalis.[48-51] The pharmacological activity of all cardiac glycosides is vested in the aglycone. The sugar moeity probably only influences lipid solubility, and thus absorption. The stereospecificity essential for digitalis-like cardio-activity is present in both the cardenolides (with a 5-membered, singly unsaturated lactone ring at position C_{17} of the steroid skeleton), and bufadienolides (with a 6-membered, doubly unsaturated lactone ring in this position). Both have the stereospecific configuration essential for cardiac activity,[52] and therefore the same pharmacological and toxicological actions.[53-56] Furthermore, in the cardiac glycosides there is always a glycosidic-bound sugar moiety at position C_3 of the steroid structure, but in the toad toxins this is replaced by an ester linkage to suberyl-arginine (Fig. 1). The pharmacological receptor for all cardiac glycosides is the membrane-bound Na^+, K^+-adenosine triphosphate (Na^+, K^+-ATPase) enzyme.[57-60] Inhibition of this enzyme decreases intracellular sodium influx. Further exchange for calcium ions results in positive cardiac inotropy.[61-63]

Digitalis is used in humans and animals to treat congestive cardiac failure and to slow the ventricular heart rate in the presence of atrial fibrillation and flutter. In toxic amounts digitalis causes arrhythmias and asystole.[64] Knowledge of toad toxicity dates back to ancient times where physicians used dried toad skins to treat dropsy (oedema) and as a cardiotonic, even before digitalis was introduced.[65,66] Dried skin extracts of toads are still employed in herbal medicines today by the Chinese (known as *Ch'an Su*) and Japanese (called *Senso*)[67,68] to treat congestive heart failure.[69] Bufadienolides, like digitalis in toxic doses, initially cause bradycardia, followed by a sequential first-, second- and third-degree atrioventricular block and terminally asystole.[47,53,70] Extracardiac effects of bufadienolides also mimic digitalis toxicity. Effects include nausea, emesis, diarrhoea and a bitter taste. However, no single substance is entirely responsible for the clinical signs of toad toxicity, as the exogenous catecholamines found in toad secretions have a marked additive effect on the toxicity of bufotoxins.[71]

TOADS

Toads are the most common amphibians that man and his pets have contact with. There are 11 species of *Bufo* (Fig. 2) and one of *Schismaderma* (Fig. 3) in the South African Bufonidae. Requiring water only to breed and for their tadpole stage, they are terrestrial animals inhabiting most of the country. Toads are common in urban areas and suburban gardens. Because all *Bufo* species are poisonous to some extent and capable of causing toxicity, species identification is unnecessary. Compared with other frogs, toads are stout, slower-moving creatures with short limbs and little webbing on their feet. Their dorsal skin, with cryptic colouration, is rough with numerous wart-like dermal protuberances. Their paratoid glands are conspicuous, pitted
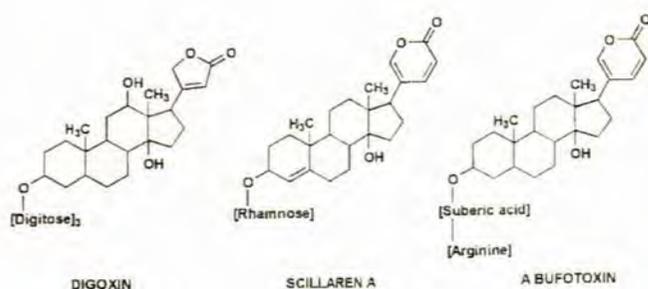
Fig. 1. *The structural formulae of the medicinal cardenolide (digoxin) and bufadienolide (scillaren A) cardiac glycosides in comparison with a typical bufadienolide toad toxin (bufotoxin).*

Fig. 2. Bufo gutturalis *(guttural toad). The paratoid glands situated behind the eyes and the wart-like dorsal dermal skin glands are very distinct (photo — V C Carruthers).*

Fig. 3. Schismaderma carens *has a conspicuous glandular ridge running dorsolaterally down each side of its body. The dorsum is brick red in colour and granular. There are no paratoid glands (photo — V C Carruthers).*

ridges located behind each eye. Toads emit secretions containing bufadienolides from these paratoid glands when frightened or injured (Fig. 4). The presence of cardiac glycoside activity in the secretions of the four largest South African *Bufo* species has been confirmed by digoxin-specific fluorescence polarisation immunoassay (T W Naudé, A R Schultz, Toxicology Division, Ondestepoort Veterinary Institute — unpublished data). Smaller amounts of poison are secreted from dorsal dermal glands, which explains why mammals that prey on toads frequently consume only the ventral parts, rejecting the dorsal skin.[72] Paratoid glands are absent in *Schismaderma* (the red toad). The dorsal skin in this species is

Fig. 4. Bufo rangeri *(raucous toad) secreting sticky white toxins from its paratoid glands (photo — W R Branch).*

usually brick-red in colour and lacks the large warts of other toads. The same type of toxins, however, are still secreted from their dorsal skin glands (T W Naudé, A R Schultz — unpublished data).

The mouthing of South African toads by dogs is known to cause intoxication, often with fatal outcome.[73-74] The development of clinical signs varies according to the victim's age, concurrent disease, amount of toxin absorbed in relation to body weight and the length of time following exposure.[25] Because toxins are rapidly absorbed through buccal and gastric mucosa, clinical symptoms are usually evident within a short period of contact with the toad.[75] Distasteful poisons cause a burning sensation and inflammation of the buccal and pharyngeal mucosa, forcing the animal to drop the toad. In cases of mild poisoning no other toxic effects occur.[76] Profuse salivation (which helps remove toxins), head shaking, vomiting, abdominal pains, diarrhoea (which may be haemorrhagic), pyrexia, laboured respiration (with possible pulmonary oedema), cyanosis and cardiac arrhythmias are cardinal clinical signs of systemic toxicity in dogs and cats. Neurological effects, including ataxia and seizures, occur with more severe poisoning.[77] Seizure activity is usually a precursor to death. Death in domestic animals has been known to occur within 15 minutes of the onset of symptoms, and in most cases ventricular fibrillation is the cause of death.[47] Toxicity may persist for a week, resulting in exhaustion of the animal.[77]

Toad toxicity in humans is less common. Ingestion of toad poisons by humans also causes local oral irritation with profuse salivation, vomiting, dizziness, blurred vision, general weakness, cyanosis, mental confusion and hallucinations, peri-oral numbness, seizures, transient focal neurological signs and metabolic disturbances.[78-80] It is unclear whether convulsions are due to direct stimulation of the central nervous system[81] or to a secondary mechanism such as arrhythmia. Cardiac manifestations of toad toxicity in humans include severe bradycardia, both hypertension and hypotension, arrhythmias and cardiopulmonary arrest. Electrocardiographic features of bufotoxins on the mammalian heart include bradycardia, P-R prolongation, T-wave inversion, ectopic beats, secondary ventricular tachycardia and ventricular fibrillation.[46,70]

Evidently the most important toxic effects in terms of risk to the patient are those involving the heart. Hence patients with premorbid cardiac disease are at increased risk. Other factors likely to increase sensitivity to bufadienolides are poor excretion of toxin (renal disease) and electrolyte disturbances, particularly potassium abnormalities.[82-83] Children and immature animals, however, seem to tolerate higher concentrations of cardiac glycosides than do adults.[84-85] Toad toxicity during pregnancy does not appear to be teratogenic[86] even though components of bufadienolides have been shown to modulate cellular differentiation and angiogenesis.[87]

Significant ocular effects have occurred in humans following eye exposure to toad secretions. Pain, chemical conjunctivitis,

swelling of the eyelids, clouding of the cornea, and changes in intra-ocular pressure have all been reported after toxic secretions were inadvertently brought into contact with the eyes.[88-91] There is also a report of a toad 'squirting' venom a distance of 2 - 3 feet into the eyes of a man, resulting in temporary blindness.[92] Application of toad venom to the mammalian eye initially causes painful irritation, followed 30 - 60 minutes later by mydriasis, swelling of the eyelids and local anaesthesia. Initially conjunctival congestion is absent. Conjunctivitis rarely persists for longer than 24 hours. Venom passing via the nasolacrimal ducts into the nose and mouth can cause salivation and sneezing.[92] Systemic poisoning can also occur with the absorption of venom from the eye.[20]

RED-BANDED RUBBER FROG

Frogs of the genus *Phrynomantis* (rubber frogs) are found on the African continent south of the Sahara. Their skin is smooth and rubbery to the touch. In South Africa, only skin secretions of the red-banded rubber frog *Phrynomantis bifasciatus* (formerly *Phrynomerus bifasciatus*) are toxic. The dorsum of this frog has aposematic (warning) red or pink markings and a ventral surface which is grey with white spots (Fig. 5). Although their digits have expanded terminal discs, these crevice-creepers are not arboreal in habit. Their milky secretions, produced in great quantities when disturbed, can cause both local irritation and systemic toxicity in humans.[19,93] Their secretions are also poisonous to other frog species that come in close contact with them.[9,93]

Skin irritation after handling this frog causes an erythematous rash with temporary inflammation.[8,9,94] Paraesthesia of the hands is also experienced (G van Aswegen — personal communication). More prolonged contact with the frog results in throbbing pain which may persist for up to 10 hours.[95] No fatalities are recorded in the literature. Only one serious toxic reaction attributed to *P. bifasciatus* has been documented.[96] Thirty minutes after contact with the toxin the scratches on the victim's hand became painfully inflamed and oedematous. One hour later he developed systemic symptoms. Painless muscle contractions of the chest and abdomen resulted in respiratory difficulty and inability to stand. One-and-a-half hours later the victim experienced a headache, dizziness, tachycardia, diaphoresis and nausea. Four hours after initial contact, toxicity appeared to subside gradually, except for a persistent myalgia. An intense burning sensation of the eyes also occurred following contact with secretions. The severity of this reaction, described by the author as an anaphylactoid reaction,[96] was thought to be related to the absorption of toxic secretions directly into the bloodstream through the open scratches on the victim's skin.

The symptoms in the aforementioned case are not typical of anaphylaxis, although humans do vary in their clinical expression of anaphylaxis.[97] Clinical manifestations of

Fig. 5. *(Top)* Phrynomantis bifasciatus *is readily identified by its striking red to pink dorsal markings against a black background. Adults are usually 5 cm in size. (Bottom) The ventral surface is grey with white spots (photos —* V C Carruthers).

anaphylaxis usually start shortly after allergen administration, and the most common manifestations are usually dermatological. More serious reactions involve the respiratory and cardiovascular systems. It is surprising that the victim in this case did not experience any significant cardiorespiratory symptoms, as skin secretions of the red-banded rubber frog are known to be cardiotoxic.[98] Biochemical analysis of *P. bifasciatus* skin secretions, however, has failed to demonstrate any identifiable active toxins (J Visser — personal communication). This frog is probably of little potential danger to humans; firstly because people are unlikely to come into contact with this nocturnal species, which spends much of its time in concealed places, and secondly because knowledgeable handlers with many years' experience appear never to have suffered any ill effects while handling specimens (N I Passmore, J Visser — personal communication).

MANAGEMENT OF INTOXICATION

General and supportive measures

To prevent possible reactions, one should avoid direct contact while handling poisonous specimens. Handlers should keep hands away from their eyes, noses and mouths. Treatment of intoxication depends on correct diagnosis. This is often difficult, particularly with paediatric and veterinary victims where a history is unreliable and many other conditions could be confused with poisoning. After suspected poisoning, urgent medical (or veterinary) attention should be sought as mortality may occur within as little as 15 minutes from time of exposure in dogs. Monitoring of the patient for 12 - 24 hours may be necessary, as delayed tissue distribution occurs with cardiac glycosides.[85] The principles of treatment in veterinary and human patients are unlikely to differ. In fact, the pharmacokinetics of cardiac glycosides in normal human subjects is similar to that in canines.[99-100]

Local skin irritation is self-limiting and unlikely to require topical antihistamines, hydrocortisone and analgesics. Applying cold ice-packs offers some relief from pain.[95] Vigorous oral irrigation and a dilute sodium bicarbonate mouthwash may help to relieve local oral symptoms.[77] If eyes become exposed to secretions immediate copious irrigation with clean water or saline is essential. Because the eye tends to clear up after 24 hours no additional treatment is required, unless infection or complications develop. Inducing emesis[101] is probably of little value once signs of toxicity are evident, as vomiting commonly occurs as a result of toad poisoning. Gastric lavage, activated charcoal and cathartics have also been used to limit the absorption of ingested toad toxins.[3] True anaphylaxis, although unlikely, will need to be treated with respiratory support, adrenaline and intravenous fluids. Parenteral fluids will also be required to treat shock and excessive vomiting, in volumes sufficient to guarantee an optional diuresis. Additional therapies include systemic corticosteroids and antihistamines,[77] even though immune reactions are insignificant.[71] Sedatives, tranquillisers and systemic analgesics are of no proven benefit.[71] Treatment with magnesium salts plus agents that chelate calcium, such as ethylenediaminetetra-acetic acid (EDTA), might decrease the toxicity of bufadienolides.[71] Convulsions can be treated with diazepam and pentobarbitone. Pentobarbital-induced anaesthesia has been shown to increase canine tolerance to *Bufo* intoxication.[25] Although unlikely, intubation and ventilation may also be necessary.[20] Fortunately, in many cases recovery usually occurs without any therapeutic assistance.

Cardiac management

The most toxic aspects of toad poisoning are the cardiac effects. A baseline ECG may help in evaluating the patient's condition by detecting treatable cardiac abnormalities. Continuous ECG monitoring will also help to establish a response to treatment. Blood chemistry, in particular potassium levels, is also important in assessing severity of intoxication. Hyperkalaemia is often suggestive of acute ingestion of cardiac glycosides.[102-103] Because the chemical structure of bufadienolides is similar to that of digoxin, toad toxins will frequently cross-react with digoxin antibodies used in digoxin immunoassays.[104]

In cases of systemic envenomation, or when the diagnosis of toad intoxication is in doubt, actual measurement of serum cardiac glycoside levels by immunoassay is required. While competitive radio-immunoassays with antibodies of broad specificity to cardiac glycosides have proved successful in confirming the diagnosis of poisoning from the cane toad *Bufo marinus*,[105] commercially available monoclonal digoxin immunoassays appear to be of little help.[106] Nevertheless there is enough cross-reactivity between digoxin and bufotoxins to qualitatively confirm cardiac glycoside activity by digoxin-specific fluorescence polarisation immunoassay in skin secretions of toads, and in the organs of dogs that died from intoxication (T W Naudé, A R Schultz — unpublished results). These immunoassays will also permit the rapid screening of other anuran species suspected of containing cardiac glycosides.

Sympatholytics, acting on both α- and β-adrenergic receptors, are effective in the treatment of toad poisoning.[71] If life-threatening arrhythmias are present, they will respond to appropriate medications. Various drugs advocated include atropine, propranolol, diphenylhydantoin, lidocaine and calcium gluconate.[25,71,107-108] Although atropine treats bradycardia, heart block and hypotension from *Bufo* intoxication, the sialogogic effects of toad poisoning do not appear to be appreciably influenced by its administration. A temporary cardiac pacemaker in cases of persistent symptomatic bradycardia may be required.[96] Working on the treatment of dogs poisoned with the cardiac glycoside and catecholamine-containing skin secretions of *Bufo marinus*, researchers found that total adrenergic blockade with propranolol and dibenamine protected animals against the toxic effects of up to 100 times the lethal dose of crude toad toxins.[71] The possibility of using acetyl promazine (as an α-blocker) in conjunction with propranolol (as a β-blocker) as a combination therapy seems worthwhile investigating. Propranolol is thought to antagonise the stimulatory effect of glycosides on sympathetic nerves, thus blocking the release of endogenous catecholamines. This action then limits the intoxication to a purely cardiac glycoside intoxication.[109] Propranolol also has anti-arrhythmic properties, not directly related to its β-receptor-blocking action.[109] Administering potassium in the presence of hypokalaemia will further antagonise cardiotoxic effects. Potassium, however, should be avoided if there is heart block. There is currently no known antidote to toad toxins. The administration of commercially available digoxin-specific antibodies may antagonise some of the cardiotoxic effects of toad venoms.[110-111] However, more work in this field is still required before the use of antibodies becomes an accepted form of treatment.

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